Selection of phage-displayed peptides on live adherent cells in microfluidic channels.

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Public Summary:
Affinity reagents that bind to specific molecular targets are an essential tool for both diagnostics and targeted therapeutics. There is a particular need for advanced technologies for the generation of reagents that specifically target cell-surface markers, because transmembrane proteins are notoriously difficult to express in recombinant form. We have previously shown that microfluidics offers many advantages for generating affinity reagents against purified protein targets, and we have now significantly extended this approach to achieve successful in vitro selection of phage-displayed peptides that recognize markers expressed on live, adherent cells within a microfluidic channel. We show that, compared to conventional biopanning methods, microfluidic selection enables more efficient discovery of peptides with higher affinity and specificity by providing controllable and reproducible means for applying stringent selection conditions against minimal amounts of target cells without loss.

Scientific Abstract:
Affinity reagents that bind to specific molecular targets are an essential tool for both diagnostics and targeted therapeutics. There is a particular need for advanced technologies for the generation of reagents that specifically target cell-surface markers, because transmembrane proteins are notoriously difficult to express in recombinant form. We have previously shown that microfluidics offers many advantages for generating affinity reagents against purified protein targets, and we have now significantly extended this approach to achieve successful in vitro selection of T7 phage-displayed peptides that recognize markers expressed on live, adherent cells within a microfluidic channel. As a model, we have targeted neuropilin-1 (NRP-1), a membrane-bound receptor expressed at the surface of human prostate carcinoma cells that plays central roles in angiogenesis, cell migration, and invasion. We show that, compared to conventional biopanning methods, microfluidic selection enables more efficient discovery of peptides with higher affinity and specificity by providing controllable and reproducible means for applying stringent selection conditions against minimal amounts of target cells without loss. Using our microfluidic system, we isolate peptide sequences with superior binding affinity and specificity relative to the well known NRP-1-binding RPARPAR peptide. As such microfluidic systems can be used with a wide range of biocombinatorial libraries and tissue types, we believe that our method represents an effective approach toward efficient biomarker discovery from patient samples.

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